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Effect of organic matter on the availability of the fixed phosphates in the soil and phosphates in rock phosphate.

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EFFECT of ORGANIC MATTER on the AVAILABILITY OF THE FIXED PHOSPHATES IN THE SOIL AND PHOSPHATES IN ROCK PHOSPHATE

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INTRODUCTION

According to the Soil Science Society's Committee on Terminology (19) fixed phosphorus is that phosphorus that has changed to less soluble forms as a result of reaction with the soil. More specifically, it is that quantity of soluble phosphorus compounds which, when added to soil, becomes chemically or biologically attached to the solid phase of soil so as not to be recovered by extracting the soil with a specified extractant under specified conditions. Phosphate fixation has been a major concern of agronomic investigators for some time. It is economically important to the farmer who adds soluble phosphates to the soil in the form of expensive fertilizers because only a small percentage remains effective for crop production - the remainder becomes fixed. A means of preventing this fixation or of releasing that which is already fixed in the soil would be of utmost value to him and to the population as a whole.

The prevention of this fixation in the soil is partially effected by banding, pelleting of phosphate fertilizers and adding phosphate fertilizers with manures. Several investigators (2,3,13,28,37,38) have reported that iron and aluminum are largely responsible for the fixation of phosphates in the neutral and acid soils. Phosphorus fixation by anion exchange within the crystalline makeup of the clay has also been proposed (22,29,35) as a mechanism of fixation. It seems well established that at pH 7.5 and above fixation is caused by calcium, magnesium, manganese, and other divalent ions.

Other investigators (1,17,20,23,39,40) believe that organic matter has an effect upon the release of those fixed soil phosphates and phosphates in rock phosphate. Swenson, Cole, and Sieling (37) Struthers and Sieling (36) have indicated that organic acids with the ability to form complexes with iron and aluminum are effective in preventing the fixation of soluble phosphates and in the releasing of that already fixed by those ions. They studied definite organic acids as to their ability to prevent the precipitation of phosphorus by iron and aluminum under controlled conditions and found that the most effective acids were citric, oxalic, tartaric, malonic, malic, and lactic. Since these and many other organic acids occur in soils as a result of the action of microorganisms on organic matter it suggested that they might have an important function in making soluble the phosphate which had been held in the soil in an insoluble form. It would be of practical significance to determine whether organic matter, when added to a soil low in available phosphates, would increase the availability of the phosphates to growing plants.

It was the purpose of this investigation to determine the effectiveness in preventing phosphate precipitation by iron of organic compounds, some of which occur in soils in appreciable quantities and which have not been studied previously. It was also the purpose to determine whether certain organic substances, which serve as readily available energy sources for microorganisms but which contain no phosphate, would have any effect on the phosphate availability of soils to which they were added.

EXPERIMENTAL

Effect of Organic Substances On Phosphate Precipitation By Iron As Influenced By pH

Several organic substances, principally hydroxy organic acids, have been found to vary in their effectiveness in preventing the precipitation of soluble phosphates by iron or aluminum. Each organic acid has its maximum effect within a rather narrow range of pH values but may be quite effective throughout a rather wide range of pH values. Some acids were most effective at very low pH values, some in moderately acid solutions, and others near the neutral point. Therefore, within a soil containing organic matter, it seems likely that some organic acid will be produced by microorganisms which will be effective in releasing the phosphate from the insoluble iron and aluminum phosphates whatever the pH value of the soil.

Soil organic matter is the product of decomposition of plant and animal residues by various microorganisms. It contains the degradation and resynthesized organic substances of the various biochemical reactions. Among the substances which occur in soils, rather large amounts are the pectins or polyuronides. Some soils are known to contain as much as 35 per cent of their organic matter in this form. Since these polyuronides contain several hydroxy acid groups in their rather complex molecules, it seemed likely that they might show the property of freeing phosphate from its insoluble form as iron and aluminum phosphates. The purpose of the following experiment was to determine the effect of pectin and galacturonic acid on the precipitation of phosphorus by iron at various pH values.

Establishment of standard values of phosphate precipitation by iron at various pH levels. A procedure similar to that used by Struthers and Sieling (36) was used in this study. A solution of 1 millimol of $FeCl_3$ containing a known amount of excess HCl was mixed with a solution containing 1 millimol of KH_2PO_4 in a 250 ml beaker. Water was added to this solution to make a final volume of 125 ml less the predetermined amount of NaOH that was required to give the desired pH value. Enough NaCl was added to make the final concentration 1 per cent. The mixture was heated to boiling and sufficient 0.2 N NaOH (carbonate free) was added to adjust the equilibrium pH to the approximate value desired. The resulting mixture was placed on the steam bath for one-half hour, cooled to room temperature for pH determination, reheated, and filtered while hot through Whatman No. 42 paper. The filtrate was received in a 500 ml volumetric flask. The precipitate was washed three times by filling the filter paper with hot 1 per cent NaCl solution adjusted approximately to the respective pH values. The volumetric flask was then filled to volume and an appropriate aliquot was used to determine the phosphate content by the ammonium molybdate method of Sherman (33). The phosphate in the filtrate was used as a measure of that not combined by an insoluble form by the iron.

The values were determined for the phosphate not precipitated by iron from pH 3 to pH 8. These values were used throughout the study as standards for the amount of phosphate not precipitated under fixed experimental conditions.

Determination of the effect of organic anions on the prevention of precipitation of phosphate at the various pH levels. The procedure used to determine the effect of the organic anions was essentially the same as the one mentioned above except that 1 millimol of the anion being considered was mixed with the phosphate solution before the iron and aluminum solutions were added. This method of procedure was also modified to get further information on the establishment of equilibrium in the systems where citric acid was the anion being studied. Curve a in Figure 1 shows the influence of pH on the effectiveness of citric acid in preventing phosphorus precipitation by iron when treated in the above manner.

The first modification was to change the sequence of adding the iron to the system. The citrate and phosphate anions were added to the beaker. The volume was made to 125 ml less the amount of NaOH to be added; then the predetermined amounts of NaOH were added to give the desired pH values. The solution was then boiled slowly for one-half hour after which time the iron was added while boiling and stirring the mixture. The mixture was allowed to cool to room temperature for pH determination, reheated and filtered while hot through Whatman No. 42 paper and the phosphorus determined as mentioned above. The curve b in Figure 1 shows greater effectiveness for the citric acid to prevent phosphate precipitation over a wider pH range than in the previous procedure and actually there was a 100 per cent

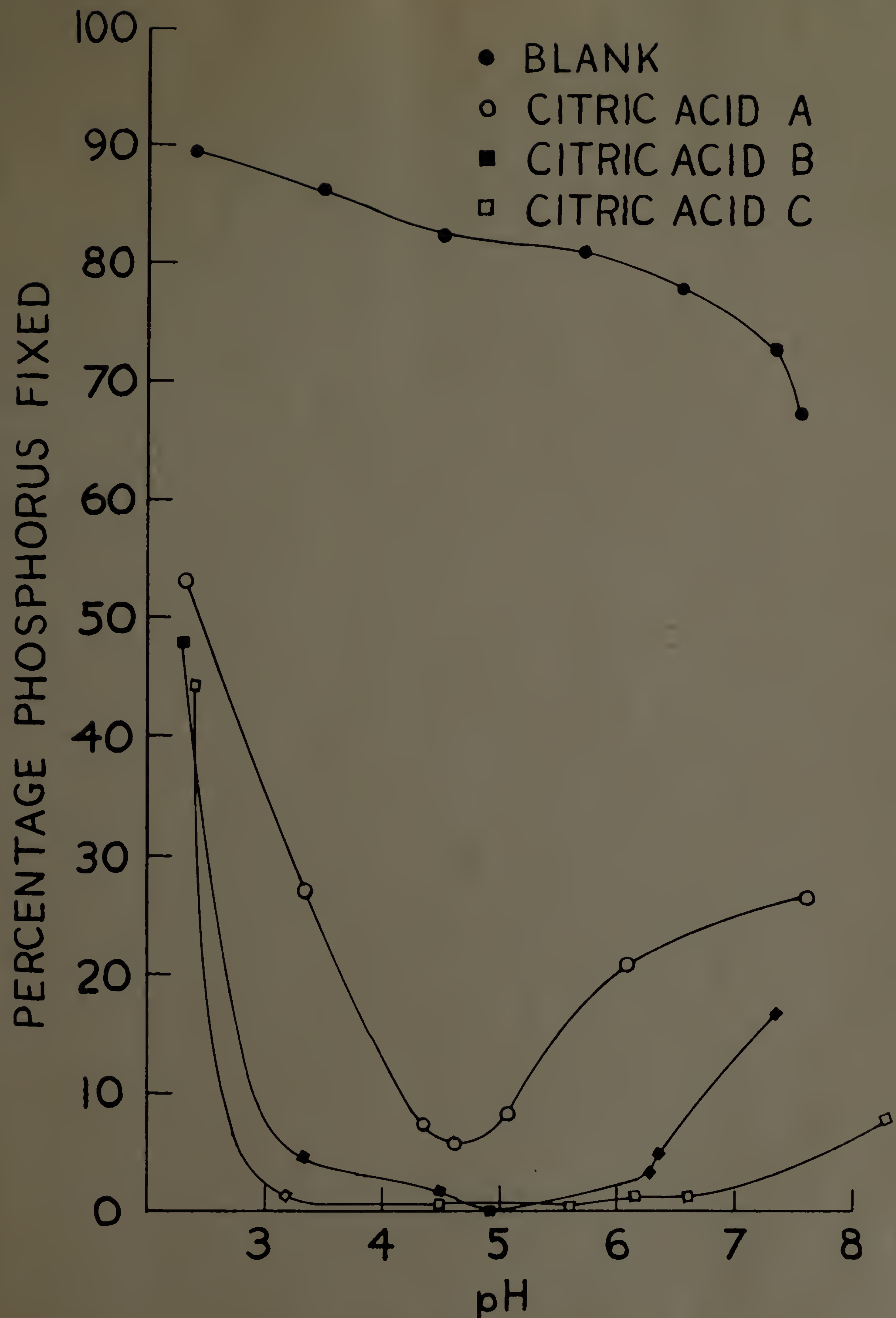

Fig. 1. Influence of pH on the Effectiveness of Citric Acid in Preventing Phosphorus Precipitation By Iron Under Different Conditions

prevention of precipitation. These results show that when the anions were in equal competition for the iron, the citrate anion had the greater attraction.

The second modification was the initial pH and the temperature of the solution at the time of the addition of the iron. The citrate and phosphate anions were added to the beaker. Water was added to the mixture to make a volume of 125 ml less that volume of NaOH to give the desired pH values. NaOH was added to give the desired pH values up to pH 4.5 where maximum effectiveness had been found to exist. To this mixture at room temperature the iron solution was added and stirred thoroughly. Enough additional NaOH was added to give the desired higher pH values. The effectiveness was shown over an even wider pH range than the previous procedure. These results show the need for a standard procedure in determining the effectiveness of the organic anions.

The organic acids studied were citric, _dl_-phenylalanine, _α d_-galacturonic acid, and citrus pectin in a 0.2N NaOH solution. The curves in Figure 2 show that citric acid was the most effective of those materials in preventing phosphorus precipitation by iron. The cyclic amino acid, phenylalanine, has been reported to be an excellent plant growth-substance former (20) but it was not effective in preventing the precipitation of phosphate by iron. Actually more phosphate was precipitated when it was present than in its absence. The _αd_-galacturonic acid, a decomposition product of pectin in nature, proved to be quite effective in preventing the phosphorus precipitation by

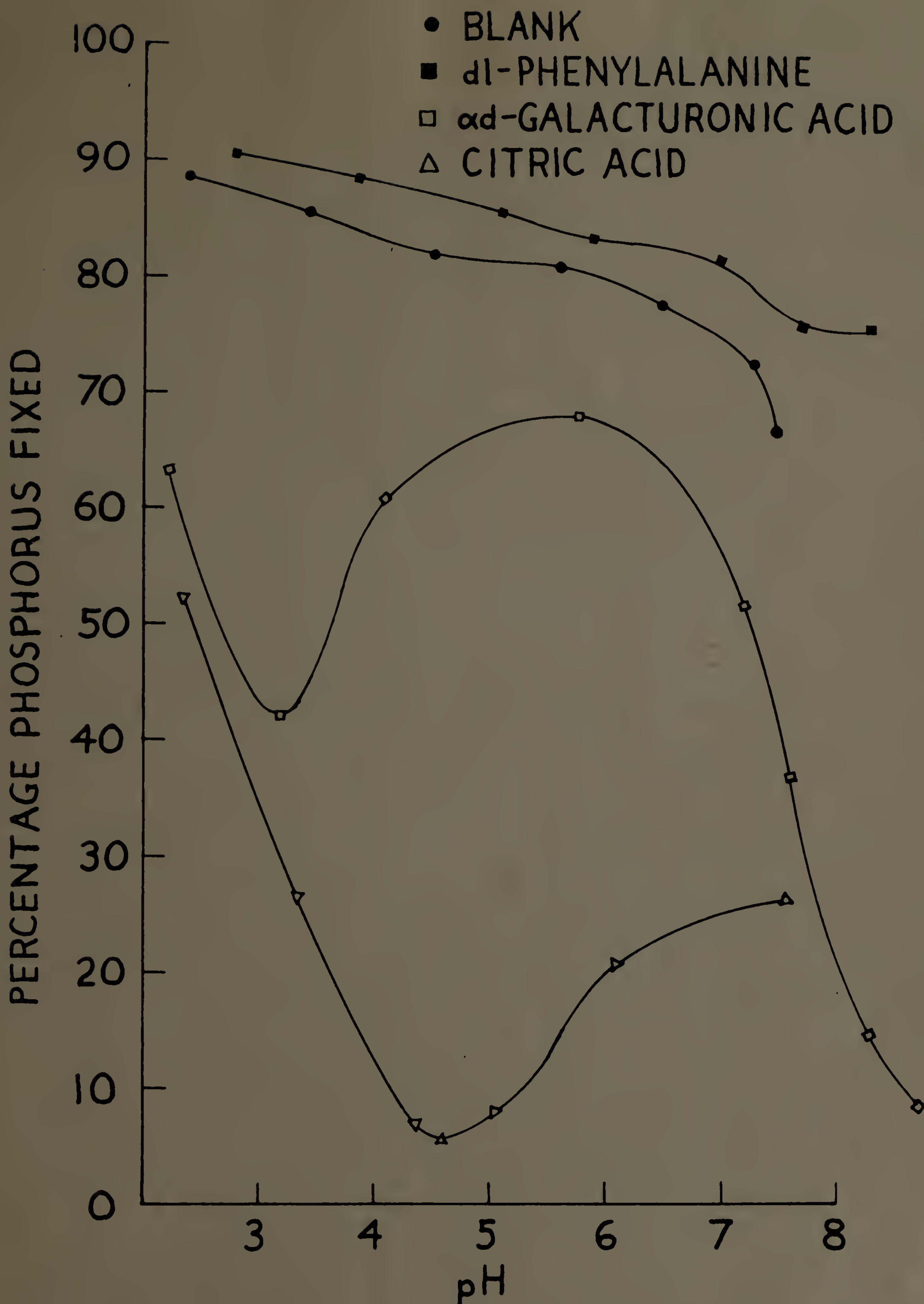

Fig. 2. Influence of pH on the Effectiveness of Organic Compounds in Preventing Phosphorus Precipitation By Iron

iron; although, less effective than citric acid up to pH 7.5. The maximum effect was at pH 3.2 and apparently pH 9 and a minimum effect at about pH 2 and pH 5.8.

A systematic method was not followed for the study of the citrus pectin solution. Preliminary studies did indicate, however, that the citrus pectin in 0.2N NaOH was a very effective agent in preventing phosphate fixation. Waksman (39) stated that uronic acids with the general formula $C_6H_{10}O_7$ generally occur as polyuronides among plant products in the form of pectin, gums, mucilages, and are especially abundant in microbial gums and slimes. This introduced the question as to whether these polyuronides decomposed and the uronic acid was able to release some of the soil phosphates. A chemical analysis by a modified method of Dickson _et al_ (8) and Pierce and Hasnisch (30) indicated that the citrus pectin used in the present study was 54.48 per cent uronic acid anhydride.

Galacturonic acid and pectin proved to be effective in preventing phosphate precipitation by iron. When one considers that these compounds are found in the organic matter of soils in varying amounts, it seems reasonable to believe that they are able to prevent the precipitation of soluble phosphates or release that phosphate already precipitated in the soil. This suggested the use of plant cultures growing in soil to study the problem. The organic matter decomposition and resulting production of organic acids is a slow but steady process as it naturally occurs; therefore, the uptake of a growing plant seemed to be the most logical tool for this type of study. Starch, citrus pectin, sucrose, and glucose were used as sources of organic matter as discussed later.

Greenhouse Studies Using Pot Cultures to Indicate Phosphate Removal From the Soil

Previous investigators have studied the effect of organic matter upon the soil phosphates. Jensen (20) reported evidence of making soil phosphates more soluble by the addition of freshly decomposing organic matter. Bauer (1) found increased phosphate uptake where mixtures of organic matter and rock phosphate were used. These investigators used organic matter which contained phosphorus. Thus resulted their inability to be certain as to the source of the increased phosphate that the plants removed. Gerretsen (17) found results which pointed to the direction that microorganisms were able to make hitherto inaccessible phosphates available to the plant. His work with plants in sterile cultures indicated that the substances excreted by the roots make $Ca_3(PO_4)_2$ and bone meal available to some extent. Copeland and Merkle (7) found that the actively decomposing organic matter was more important in increasing the phosphate uptake by plants than was the total organic matter content of the soil. Waksman (39) stated that humus exerted a definite effect upon the availability of the inorganic phosphorus in the soil and that rock phosphate interacted with some of the humus constituents to render some of the phosphate more soluble. In a study of this type it seemed highly desirable to use a source of organic matter which contained no phosphorus. This would leave no doubt as to the source of the phosphates removed by the plant.

<u>Influence of organic matter upon the uptake of phosphates by corn</u>. The soil used for this study was a Merrimac fine sandy loam of pH 5.2. Swenson, Cole, and Sieling (37) showed that approximately 90 per cent of the phosphate was still fixed by iron and aluminum at pH 6.5, and that progressively more was fixed at lower pH values. Therefore a soil at pH 5.2 would be quite effective in phosphate fixation. A medium phosphate reading was obtained by the Morgan Universal System (24) or 0.002 millimol phosphorus per 100 gm. of soil. By the Bass and Sieling method (2) the phosphate fixing capacity of the soil was 30.99 millimols phosphorus per 100 gm. of soil and the citric acid soluble phosphate was 2.62 millimols phosphorus per 100 gm. of soil which represented 8.45 percentage saturation to its phosphate fixing capacity.

The soil was brought from the field and thoroughly mixed. On the oven dry basis 25 pounds of soil were added to each glazed three gallon jar as the respective treatments were mixed with the soil. Three replicates of each treatment were used.

Check treatments were no-phosphate, 80 pounds P_2O_5 per acre, and 160 pounds P_2O_5 per acre added as KH_2PO_4. The variable treatments were no-phosphate plus the respective organic matter and 80 pounds P_2O_5 per acre plus the respective organic matter. Check treatments with rock phosphate* were 80, 160, and 320 pounds P_2O_5 per acre and the variable treatments were 80 pounds

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*The term "rock phosphate" applies to pulverized Tennessee phosphate.

and 160 pounds P_2O_5 per acre plus the respective organic matter. Sucrose, glucose, starch, and citrus pectin at various rates of applications were used. Sucrose was added at 1 and 2 ton rates of application. Glucose and starch respectively were added at the rate of 2 tons per acre and citrus pectin at the rate of 1 ton per acre. Chemical analyses were made to determine the phosphate content of the organic materials that were to be added. Sucrose and glucose contained no phosphorus. Phosphorus was added in the starch at the rate of 0.54 pounds P_2O_5 per acre, in the citrus pectin at the rate of 0.51 pounds P_2O_5 per acre, and in the 3 kernels of corn at the rate of 0.242 pounds P_2O_5 per acre for Quebec 83_{10} and 0.298 pounds per acre for Quebec 83_{11} inbred lines. These were amounts so small that it is reasonable to believe that they had no significant effect upon the results.

The corn was seeded July 16, 1949 and allowed to grow until it was cut on August 3, 1949 at the tasseling stage. The plant material was dried in a steam drier for two days and was weighed, then ground in a Wiley mill. Moisture content of the plant material was determined and then the phosphorus content was determined.

The phosphorus content was determined by using a one gram sample of oven dry plant material. Duplicate samples were analyzed. The sample was placed in a tall 250 ml beaker and 10 ml of 1:1 HNO_3 solution were added. This was placed on the steam bath for one-half hour or until the organic matter was gelatinous. To the beaker 5 ml of concentrated HNO_3 plus 7 ml of 60 per cent $HClO_4$ were added. The material was digested on the hot plate until the solution was clear and dense white fumes of $HClO_4$ persisted, then continued for 20 minutes longer. The solution was cooled, diluted with hot water and transferred through Whatman 41-H filter paper into a 250 ml volumetric flask. A suitable aliquot was taken for phosphorus determination by the ammonium molybdate method of Sherman (33) modified for plant material. The total removal of phosphate from each pot was calculated from the percentages and oven dry yields of the plant material from each jar. The data in Table 1 show the soil treatments for the corn study. To eliminate any other limiting factors except phosphate the rates of 150 pounds of nitrogen per acre in the forms of $Ca(NO_3)_2 \cdot 4H_2O$ and $Mg(NO_3)_2 \cdot 6H_2O$ and potassium of 100 pounds of K_2O per acre in the form of KCl were added.

The moisture content of the soil was adjusted to approximately 20 per cent. To each jar 4 kernels of corn were planted and thinned to three plants after they were approximately three inches high. Due to an unforeseen shortage of corn kernels two inbred lines of corn were used. The two inbred lines of corn were Quebec 83_{10} and Quebec 83_{11} which were developed at the Massachusetts Station. These lines developed purple lower leaves at low phosphate levels.

Table 1. Soil treatments and varieties used for the greenhouse study of phosphorus uptake by corn

No. of Treat- ment	Variety*	Amount of compounds and organic constituent added per 25 lbs. soil (grams)								
		$Ca(NO_3)_2 \cdot 4H_2O$	$Mg(NO_3)_2 \cdot 6H_2O$	KH_2PO_4	Raw Rock Phosphate	KCl	Cane Sugar	Corn Starch	Citrus Pectin	Dextrose
I	$Q83_{10}$	3.588	3.832			0.9496				
II	$Q83_{10}$	3.588	3.832	0.8698		0.4731				
III	$Q83_{10}$	3.588	3.832	1.7396						
IV	$Q83_{11}$	3.588	3.832			0.9496	11.34			
V	$Q83_{11}$	3.588	3.832	0.8698		0.4731	11.34			
VI	$Q83_{11}$	3.588	3.832			0.9496	22.68			
VII	$Q83_{11}$	3.588	3.832	0.8698		0.4731	22.68			
VIII	$Q83_{10}$	3.588	3.832			0.9496		22.68		
IX	$Q83_{10}$	3.588	3.832	0.8698		0.4731		22.68		
X	$Q83_{10}$	3.588	3.832			0.9496			11.34	
XI	$Q83_{10}$	3.588	3.832	0.8698		0.4731			11.34	
XII	$Q83_{10}$	3.588	3.832		1.6197	0.9496				
XIII	$Q83_{10}$	3.588	3.832		3.2394	0.9496				
XIV	$Q83_{10}$	3.588	3.832		6.4788	0.9496				
XV	$Q83_{10}$	3.588	3.832		1.6197	0.9496	11.34			
XVI	$Q83_{10}$	3.588	3.832		3.2394	0.9496	11.34			
XVII	$Q83_{10}$	3.588	3.832		1.6197	0.9496	22.68			
XVIII	$Q83_{10}$	3.588	3.832		3.2394	0.9496	22.68			
XIX	$Q83_{10}$	3.528	3.832		1.6197	0.9496		22.68		
XX	$Q83_{10}$	3.588	3.832		3.2394	0.9496		22.68		
XXI	$Q83_{10}$	3.588	3.832		1.6197	0.9496			11.34	
XXII	$Q83_{10}$	3.588	3.832		3.2394	0.9496			11.34	
XXIII	$Q83_{10}$	3.588	3.832			0.9496				22.68
XXIV	$Q83_{10}$	3.588	3.832	0.8698		0.4731				22.68
XXV	$Q83_{10}$	3.588	3.832		1.6197	0.9496				22.68
XXVI	$Q83_{10}$	3.588	3.832		3.2394	0.9496				22.68
XXVII	$Q83_{11}$	3.588	3.832			0.9496				

*Q indicates the inbred line Quebec.

The results as indicated by the phosphates removed by the corn are shown in Tables 2 and 3 and in Figure 3. In Table 2 the results of the checks show that the soil used was not completely depleted of available phosphates. It did, however, respond to the application of soluble phosphates and to the addition of two tons of glucose per acre. Where 80 and 160 pounds of P_2O_5 per acre were added as KH_2PO_4 the increased phosphate uptake was 26.3 per cent and 60.5 per cent respectively. In the case of the 2 tons of glucose the increase was 20.9 per cent. There appeared to be no increase in the case of the starch and pectin material. These later materials are more complex in nature; therefore, it seems logical to expect them to be more slowly decomposed by the microorganisms. Norman (25) found evidence that the uronic group is biologically less available than the hemicelluloses. It was interesting to note the greater ability of the inbred line Quebec 83_{11} to remove phosphates from the soil. Since the removal from this variety on the checks was over 216 per cent greater than the inbred line Quebec 83_{10}, it seemed of no value to compare the treatments between inbred lines. With Quebec 83_{11} it appeared as if the sucrose decreased the phosphorus uptake when compared to the check. The 80 pounds P_2O_5 per acre as KH_2PO_4 brought the uptake about equal to the check; therefore, it seems that for this short period the corn roots could not compete with the microorganisms for the available phosphates or those released by organic acids. Furthermore this was shown because in each case

Table 2. Yield of corn and phosphates removed with KH_2PO_4 as the phosphate source

No. of Treat- ment	Phosphate Added: P_2O_5 per acre	Phosphate Added: P per acre	Phosphate Added: P per 3 pots	O.M. Added: Source	O.M. Added: per acre	Removed: Plant Material*	Removed: P	Removed: %P	Variety**
	lbs.	lbs.	mgm.		tons	gm.	mgm.		
I						140.96	167.19	0.1186	$Q83_{10}$
II	80	34.93	594			182.82	211.14	0.1155	$Q83_{10}$
III	160	79.86	1188			243.51	268.63	0.1103	$Q83_{10}$
VIII				Starch	2	159.66	163.84	0.1026	$Q83_{10}$
IX	80	34.93	594	Starch	2	181.21	219.80	0.1213	$Q83_{10}$
X				Pectin	1	155.51	162.24	0.1043	$Q83_{10}$
XI	80	34.93	594	Pectin	1	175.92	193.58	0.1100	$Q83_{10}$
XXIII				Glucose	2	164.98	192.58	0.1167	$Q83_{10}$
XXIV	80	34.93	594	Glucose	2	202.34	205.30	0.1015	$Q83_{10}$
XXVII						227.95	527.74	0.2315	$Q83_{11}$
IV				Sucrose	1	209.56	430.73	0.2055	$Q83_{11}$
V	80	34.93	594	Sucrose	1	272.07	542.06	0.1992	$Q83_{11}$
VI				Sucrose	2	219.14	413.35	0.1886	$Q83_{11}$
VII	80	34.93	594	Sucrose	2	267.29	468.61	0.1753	$Q83_{11}$

* Oven dry weight is used throughout the study.
**Q indicates the inbred line Quebec.

Table 3. Yield of corn and phosphates removed with raw rock phosphate as the phosphate source

No. of Treatment	Phosphate Added: P_2O_5 per acre	Phosphate Added: P per acre	Phosphate Added: P per 3 pots	O.M. Added: Source	O.M. Added: per acre	Removed per 3 Pots: Plant Material	Removed per 3 Pots: P	Removed per 3 Pots: %P	Variety
	lbs.	lbs.	mgm.		tons	gm.	mgm.		
XII	80	34.93	594			141.62	149.97	0.1059	$Q83_{10}$
XIII	160	79.86	1188			152.38	170.82	0.1121	$Q83_{10}$
XIV	320	159.72	2376			164.42	176.10	0.1071	$Q83_{10}$
XV	80	34.93	594	Sucrose	1	159.82	186.58	0.1168	$Q83_{10}$
XVI	160	79.86	1188	Sucrose	1	147.10	172.52	0.1173	$Q83_{10}$
XVII	80	34.93	594	Sucrose	2	168.12	201.26	0.1197	$Q83_{10}$
XVIII	160	79.86	1188	Sucrose	2	160.35	174.31	0.1087	$Q83_{10}$
XIX	80	34.93	594	Starch	2	164.54	200.02	0.1216	$Q83_{10}$
XX	160	79.86	1188	Starch	2	176.68	197.01	0.1115	$Q83_{10}$
XXI	80	34.93	594	Pectin	1	163.11	177.40	0.1088	$Q83_{10}$
XXII	160	79.86	1188	Pectin	1	146.64	186.37	0.1271	$Q83_{10}$
XXV	80	34.93	594	Glucose	2	195.73	235.13	0.1201	$Q83_{10}$
XXVI	160	79.86	1188	Glucose	2	167.99	191.20	0.1138	$Q83_{10}$

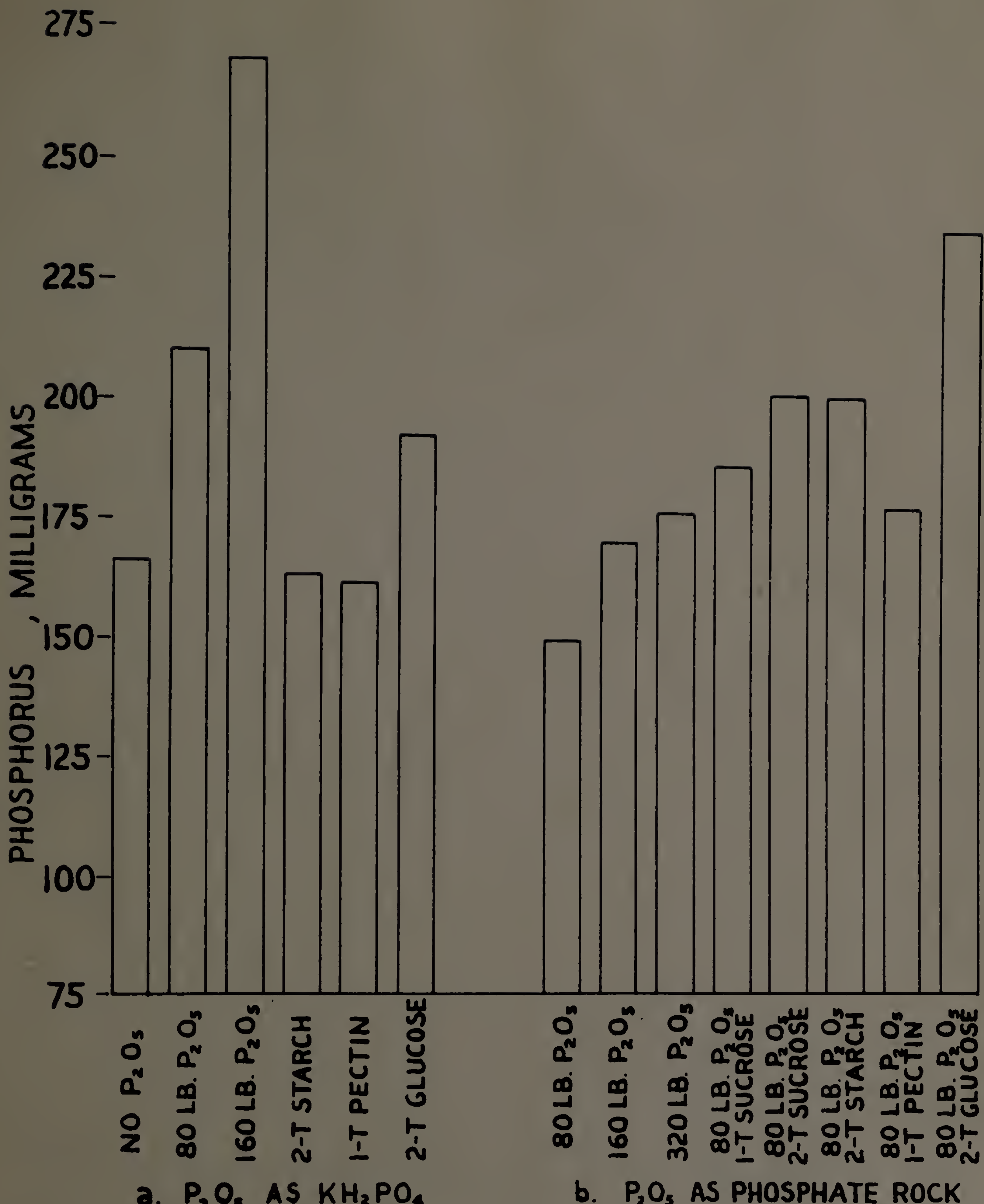

Fig. 3. Uptake of Phosphorus by Corn Showing the Effects of Organic Matter Added to the Soil

two tons of sucrose showed a decrease from one tone of sucrose. Chang (6) found that greater amounts of organic phosphorus were synthesized as the concentration of phosphate was increased.

As indicated by the data of Table 3 the soil showed no significant response to rock phosphate when added alone to the soil even at the rate of 320 pounds of P_2O_5 per acre. Slight response was shown by the addition of 1 ton of sucrose per acre and the increase was even more when 2 tons were added which in this case was about equal to that response shown by starch. The 2 tons of sucrose and 2 tons of starch approached the results obtained from 80 pounds of P_2O_5 per acre in the soluble form. The 2 tons of glucose exceeded the results obtained from adding 80 pounds P_2O_5 per acre and compared favorably with those results obtained from adding 160 pounds P_2O_5 per acre in the soluble form. It was not surprising to note that in each case where organic matter was added with rock phosphate there was an increase in phosphate uptake. Midgley and Dunklee (23) found a response in phosphate uptake where organic matter was added as manure. Waksman and Starkey (40), Shorey (34), and Forsyth (10,11,12) indicated that organic acids are produced upon the decomposition of organic matter. Swenson et al (37) showed that organic anions prevented the fixation of the phosphate ions by iron and aluminum. The organic compounds added in the present study contained no phosphorus. It seems logical to believe that upon soil microbiological activity small amounts of organic anions were produced. These acids in turn released the phosphate ions from the iron and aluminum phosphates in the soil and the calcium phosphates in the phosphate rock.

Influence of organic matter upon the uptake of phosphates by ladino clover. The soil of two replicates of each treatment from the corn study was used for the ladino clover study. The soil from the two pots was thoroughly mixed and by weight one-third of the soil was used in each of three shallow glazed steam table jars. The check soils were not treated further with KH_2PO_4 or rock phosphate. This allowed a longer time to study the effect on the soil phosphates. The variable treatments with the starch and citrus pectin were 2 and 1 tons respectively. The variable treatments with sucrose were applied periodically to make total rates of 10 and 20 tons of sucrose and with glucose at the rate of 20 tons per acre. In order to keep a more even population of microorganisms in the soil these latter organic compounds were added in increments of 1/2 and 1 ton rates per acre. Those additions of soluble organic compounds were made over a period of 20 weeks. The data in Table 4 show the soil treatments used for the greenhouse study of phosphate uptake by ladino clover.

In order to eliminate all limiting factors except phosphates the following nutrient elements in solution were added to the soil. An initial application of nitrogen was added at the rate of 75 pounds per acre as $Ca(NO_3)_2 \cdot 4H_2O$. Potassium was added at the rate of 50 pounds K_2O per acre as KCl. Magnesium was added at the rate of 64 pounds per acre as $MgSO_4 \cdot 7H_2O$. A rate of

Table 4. Soil treatments used for the greenhouse study of phosphate uptake by ladino clover

No. of Treat-ment	Amount of compounds and organic constituents added per 16.7 lbs. soil (grams)								
	*$Ca(NO_3)_2 \cdot 4H_2O$	KCl	$MgSO_4 \cdot 7H_2O$	$Na_2B_4O_7 \cdot 10H_2O$	*KNO_3	Cane Sugar	Corn Starch	Citrus Pectin	Dextrose
I	23.116	0.3607	2.288	0.1514	7.511				
II	23.116	0.3607	2.288	0.1514	7.511				
III	23.116	0.3607	2.288	0.1514	7.511				
IV	23.116	0.3607	2.288	0.1514	7.511	75.60			
V	23.116	0.3607	2.288	0.1514	7.511	75.60			
VI	23.116	0.3607	2.288	0.1514	7.511	151.20			
VII	23.116	0.3607	2.288	0.1514	7.511	151.20			
VIII	23.116	0.3607	2.288	0.1514	7.511		15.12		
IX	23.116	0.3607	2.288	0.1514	7.511		15.12		
X	23.116	0.3607	2.288	0.1514	7.511			7.56	
XI	23.116	0.3607	2.288	0.1514	7.511			7.56	
XII	23.116	0.3607	2.288	0.1514	7.511				
XIII	23.116	0.3607	2.288	0.1514	7.511				
XIV	23.116	0.3607	2.288	0.1514	7.511				
XV	23.116	0.3607	2.288	0.1514	7.511	75.60			
XVI	23.116	0.3607	2.288	0.1514	7.511	75.60			
XVII	23.116	0.3607	2.288	0.1514	7.511	151.20			
XVIII	23.116	0.3607	2.288	0.1514	7.511	151.20			
XIX	23.116	0.3607	2.288	0.1514	7.511		15.12		
XX	23.116	0.3607	2.288	0.1514	7.511		15.12		
XXI	23.116	0.3607	2.288	0.1514	7.511			7.56	
XXII	23.116	0.3607	2.288	0.1514	7.511			7.56	
XXIII	23.116	0.3607	2.288	0.1514	7.511				151.20
XXIV	23.116	0.3607	2.288	0.1514	7.511				151.20
XXV	23.116	0.3607	2.288	0.1514	7.511				151.20
XXVI	23.116	0.3607	2.288	0.1514	7.511				151.20
XXVII	23.116	0.3607	2.288	0.1514	7.511				

*Added weekly throughout the study.

25 pounds of nitrogen was added as $Ca(NO_3)_2 \cdot 4H_2O$ and as KNO_3 when nitrogen applications were made throughout the study. The idea was to force the growth of the plants by adding nitrogen, calcium, and potassium no oftener than once a week without getting toxic effects from an excess amount of salts in the soil. $Ca(NO_3)_2 \cdot 4H_2O$ was added a total of 25 times throughout the study and KNO_3 a total of 11 times. A total of 40 pounds per acre of borax were added at two separate applications.

The soil was adjusted to approximately 20 per cent moisture content and the surface allowed to dry. Eight ladino clover stolons were planted in the soil of each pot. In order to reduce the error caused by differences between plants the following method was used. Enough stolons were obtained from a single parent plant to supply a single stolon for every jar where possible or for at least one complete replicate of jars, whichever was possible. The plant material was cut for analysis from the ladino clover every six weeks for a period of 30 weeks. After that period the plant material was cut every three weeks and two of these cuttings were used for a 6-week analysis. This was necessitated by the greater amount of growth during the summer period. The study covered a period of 42 weeks of growing time for the ladino clover.

The plant material was dried in a steam oven, weighed, and ground in a micro-Wiley mill. One-gram samples from each pot were used to determine the phosphate content of the plant material. The method was the same as that previously described for the corn samples.

In Table 5 the cumulative yield results of this study are shown by 6-week periods of analysis where KH_2PO_4 was the residual phosphate source. The data show in each cutting a response in yield from the addition of soluble phosphate to the soil. No greater increase was shown between the 160 pounds and 80 pounds P_2O_5 per acre until the sixth analysis. In every case the yield was greater where starch and glucose were added to the soil. Of the two, starch produced the greater effect. Pectin showed a decrease in yield for the first four cuttings after which it showed an increase and the total yield was greater than the check.

In Table 6 the cumulative yield results are shown with rock phosphate as the residual phosphate source. The data show a significant difference in yield the fourth analysis and continued through the seventh. The total effect was no greater from 320 than from 160 pounds P_2O_5 per acre for the length of time of the study. In every case, except the starch, where the organic matter was added, the total yield was greater than the check. The order of decreasing effectiveness was 20 and 10 tons of sucrose, one ton of pectin and 20 tons of glucose.

The above data of yield do not necessarily indicate the effect of the organic matter upon soil phosphates. The results of phosphate removal from the soil where KH_2PO_4 was the residual source of phosphate are shown in Table 7 and Figures 4a and 5. The data in Table 7 show a significant increase in phosphate uptake from the residual effect of 80 and 160 pounds P_2O_5 per acre as KH_2PO_4 to the soil. Some increase was shown from the addition

Table 5. Cumulative yields of ladino clover with KH_2PO_4 as the residual phosphate source

No. of Treatment	Treatment Per Acre: Residual P_2O_5 lbs.	Treatment Per Acre: Organic Matter tons	Yield in gm. Date of Cutting: Nov. 22	Jan. 3	Feb. 14	Mar. 28	May 9	June 20	Aug. 1
I			22.31	41.54	55.42	81.13	128.20	192.04	251.58
II	80		25.82	47.99	65.19	91.70	138.08	198.87	258.87
III	160		26.00	47.16	65.16	93.59	139.85	207.34	267.44
VIII		2-Starch	38.17	64.62	86.40	116.42	167.58	234.55	299.52
IX	80	2-Starch	35.60	57.95	76.31	103.58	150.80	215.00	277.24
X		1-Pectin	18.75	36.63	51.63	78.77	128.46	192.31	256.32
XI	80	1-Pectin	22.97	43.09	59.92	88.64	139.82	205.12	267.29
XXIII		20-Glucose	28.02	47.51	64.61	94.49	145.37	213.30	281.68
XXIV	80	20-Glucose	30.91	49.40	66.03	95.04	145.09	212.47	278.42
*XXVII			28.09	47.54	64.14	90.80	139.67	202.71	262.79
IV		10-Sucrose	26.88	45.09	61.10	88.75	136.29	197.99	256.96
V	80	10-Sucrose	31.26	51.59	68.04	96.46	146.97	214.27	271.92
VI		20-Sucrose	31.50	50.09	66.50	95.28	147.63	212.77	273.71
VII	80	20-Sucrose	25.55	47.16	62.99	92.23	141.36	206.61	268.29

*Treatments XXVII, IV, V, VI, and VII are not to be considered in the study of results of other treatments as they had the residual effects of a different variety of corn.

Table 6. Cumulative yields of ladino clover with rock phosphate as the residual phosphate source

No. of Treatment	Treatment Per Acre: Residual P_2O_5 lbs.	Treatment Per Acre: Organic Matter tons	Yield in gm. Date of Cutting: Nov. 22	Jan. 3	Feb. 14	Mar. 28	May 9	June 20	Aug. 1
I			22.31	41.54	55.42	81.13	128.20	192.04	251.58
XII	80		21.57	40.79	56.08	87.13	136.16	202.36	269.99
XIII	160		17.29	33.16	48.36	76.92	130.90	206.86	282.67
XIV	320		21.42	40.04	55.72	88.81	141.48	210.65	281.93
XV	80	10-Sucrose	25.49	45.48	62.18	91.09	142.50	208.44	276.29
XVI	160	10-Sucrose	24.66	44.97	61.44	90.14	139.06	207.91	274.22
XVII	80	20-Sucrose	24.93	43.43	58.86	88.03	141.73	211.33	280.36
XVIII	160	20-Sucrose	24.22	42.11	59.14	88.88	139.71	208.64	283.06
XIX	80	2-Starch	34.70	58.29	75.75	101.62	147.84	207.61	264.49
XX	160	2-Starch	37.51	62.99	82.75	110.53	155.82	214.76	273.23
XXI	80	1-Pectin	25.04	46.11	64.47	94.44	144.00	209.94	275.72
XXII	160	1-Pectin	27.73	51.48	70.03	99.03	146.86	211.32	278.09
XXV	80	20-Glucose	29.51	49.72	66.68	94.30	141.76	207.92	274.87
XXVI	160	20-Glucose	28.61	45.97	61.08	89.99	143.26	208.27	277.23

Table 7. Cumulative phosphates removed by ladino clover with KH_2PO_4 as the residual phosphate source

No. of Treat-ment	Treatment Per Acre: Residual P_2O_5 lbs.	Treatment Per Acre: Organic Matter tons	P removed in mgm. Date of Cutting: Nov. 22	Jan. 3	Feb. 14	Mar. 28	May 9	June 20	Aug. 1
I			61.48	117.67	147.63	215.75	328.55	505.62	675.87
II	80		76.75	147.42	189.92	263.94	370.41	547.95	741.15
III	160		76.94	146.86	191.44	274.85	393.67	586.01	790.54
VIII		2-Starch	108.09	195.81	250.36	341.23	474.04	672.14	890.49
IX	80	2-Starch	102.04	170.78	214.44	292.42	411.50	603.01	821.27
X		1-Pectin	52.23	104.84	140.43	216.50	343.44	519.71	725.24
XI	80	1-Pectin	63.47	124.85	164.45	241.51	363.02	537.62	740.04
XXIII		20-Glucose	85.11	150.61	195.42	280.74	407.63	599.80	819.69
XXIV	80	20-Glucose	84.15	144.53	189.05	275.35	401.15	592.97	795.61
XXVII			80.13	140.20	179.58	254.15	375.84	562.49	761.53
IV		10-Sucrose	77.80	138.69	181.06	265.45	390.97	574.34	771.80
V	80	10-Sucrose	87.78	152.65	195.75	281.72	405.79	596.50	776.68
VI		20-Sucrose	84.51	147.85	193.79	283.36	416.04	606.16	803.75
VII	80	20-Sucrose	78.76	136.27	178.24	267.35	395.17	580.41	774.96

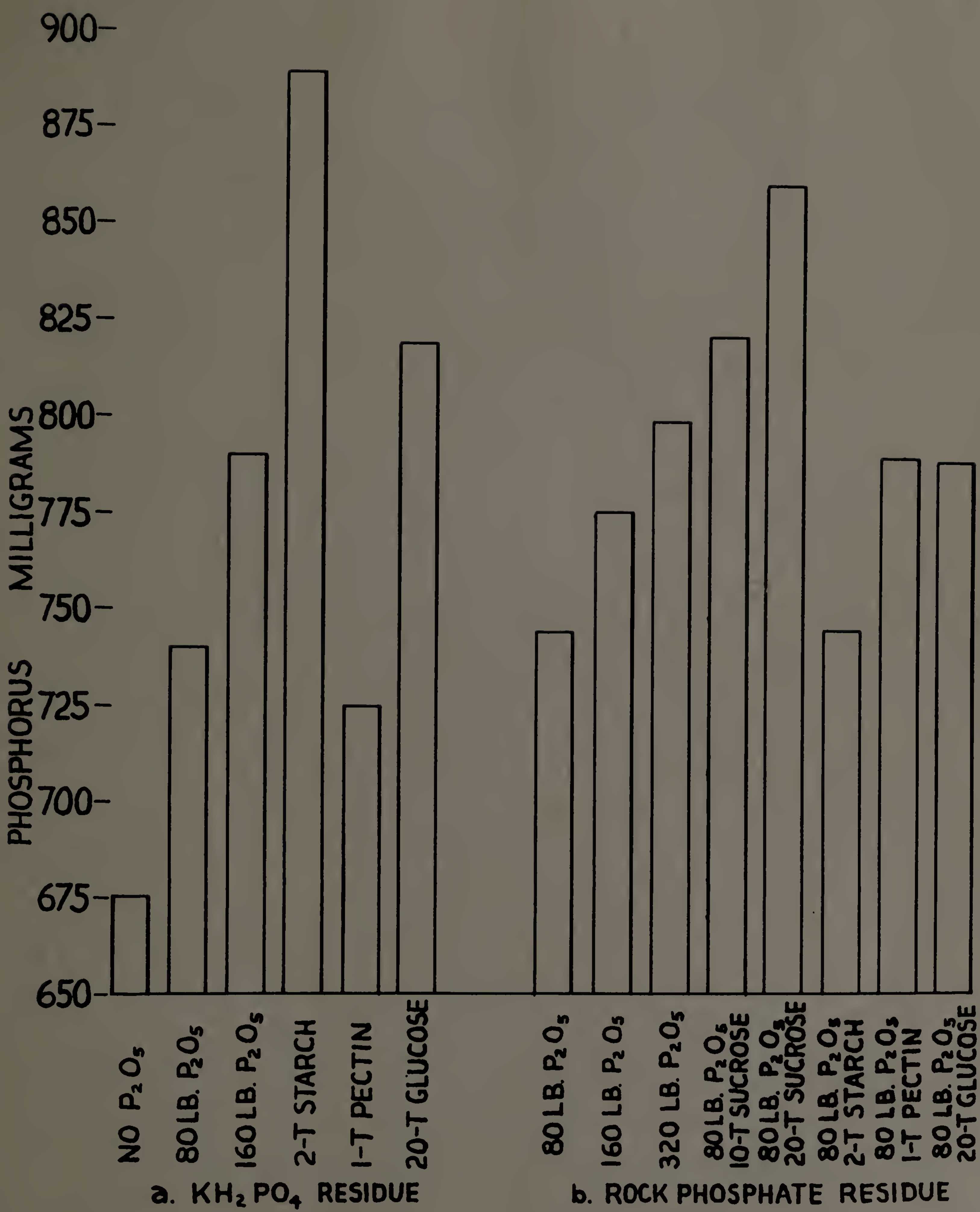

Fig. 4. Cumulative Uptake of Phosphates by Ladino Clover From Two Sources of Phosphates

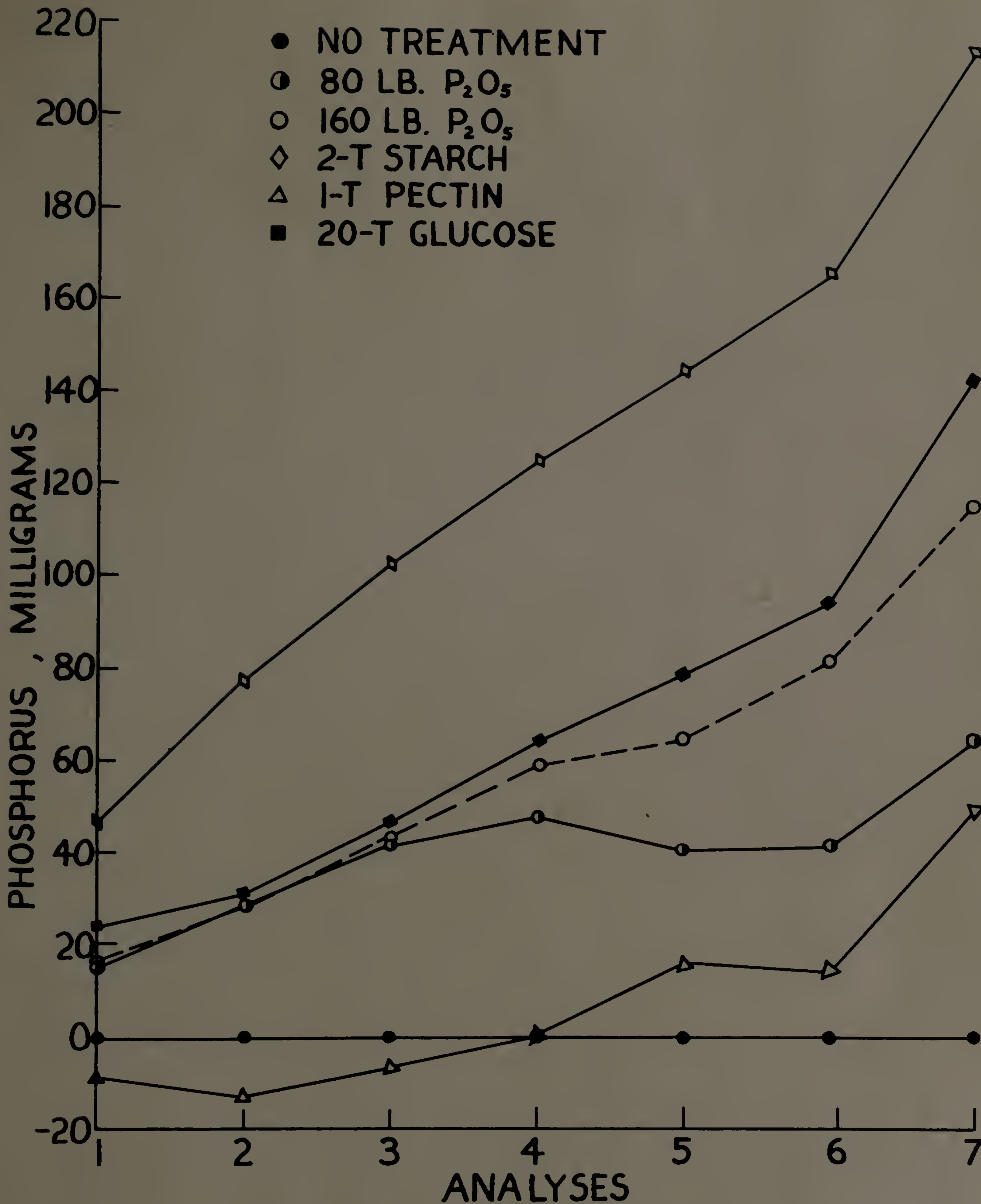

Fig. 5. Relative Uptake of Phosphates From Soils Receiving No-phosphorus, Two Applications, and Three Kinds of Organic Matter

of 1 ton per acre of pectin. Outstanding results were shown from the addition of 20 tons of glucose and 2 tons of starch per acre. Both were overwhelmingly greater in their effect upon phosphate removal than the 160 pounds P_2O_5 per acre. In Figure 4a is shown graphically the relative responses of the treatments where KH_2PO_4 was the residual source of phosphate. The data in Figure 5 show the cumulative difference of the check of phosphate removed from three replicates where KH_2PO_4 was the phosphate residue. The effectiveness of the added organic matter was shown here to be apparent throughout the seven growth periods used for analyses. Apparently the organisms that decomposed the pectin were more successful in competing with the roots for the phosphates for the first three cuttings. Apparently after the population of that particular group of organisms became great enough, then the decomposition products were sufficient to liberate soil phosphates. Apparently the decomposing organisms were able to decompose the starch and glucose more quickly as the response was shown from the first cutting. It is interesting to note that glucose and especially starch were more effective than the residual effect of the 160 pounds of P_2O_5 per acre as KH_2PO_4.

In Table 8, Figures 4b, and 6 are shown the results of organic matter upon the phosphate removal from the soil where rock phosphate was the residual source. The data in Table 8 show an increase in uptake of phosphates where rock phosphate was the residual phosphate source. In each case the addition of organic matter resulted in the increase of phosphate removed.

Table 8. Cumulative phosphates removed by ladino clover with rock phosphate as the residual phosphate source

No. of Treatment	Treatment Per Acre: Residual P_2O_5 lbs.	Treatment Per Acre: Organic Matter tons	P removed in mgm. Date of Cutting: Nov. 22	Jan. 3	Feb. 14	Mar. 28	May 9	June 20	Aug. 1
I			61.48	117.67	147.63	215.75	328.55	505.62	675.87
XII	80		61.30	119.06	153.89	237.18	355.17	534.49	744.76
XIII	160		45.85	91.67	125.52	204.78	336.31	544.59	776.82
XIV	320		62.09	119.41	157.75	250.90	385.14	576.82	799.26
XV	80	10-Sucrose	75.71	140.98	184.16	273.19	409.05	604.30	820.93
XVI	160	10-Sucrose	72.45	135.11	177.29	262.38	380.59	571.43	778.31
XVII	80	20-Sucrose	75.59	142.73	188.02	278.99	421.11	630.45	861.55
XVIII	160	20-Sucrose	74.28	136.25	186.58	279.64	419.12	631.34	880.73
XIX	80	2-Starch	95.82	168.96	210.63	285.91	396.80	560.82	744.13
XX	160	2-Starch	99.76	177.90	222.62	296.97	393.80	551.97	729.63
XXI	80	1-Pectin	73.25	142.75	193.30	276.64	397.80	586.53	790.74
XXII	160	1-Pectin	77.33	150.54	191.62	269.77	380.92	556.27	752.69
XXV	80	20-Glucose	82.41	145.66	190.05	269.59	385.12	571.17	789.93
XXVI	160	20-Glucose	82.64	138.55	177.08	260.63	392.93	577.99	794.85

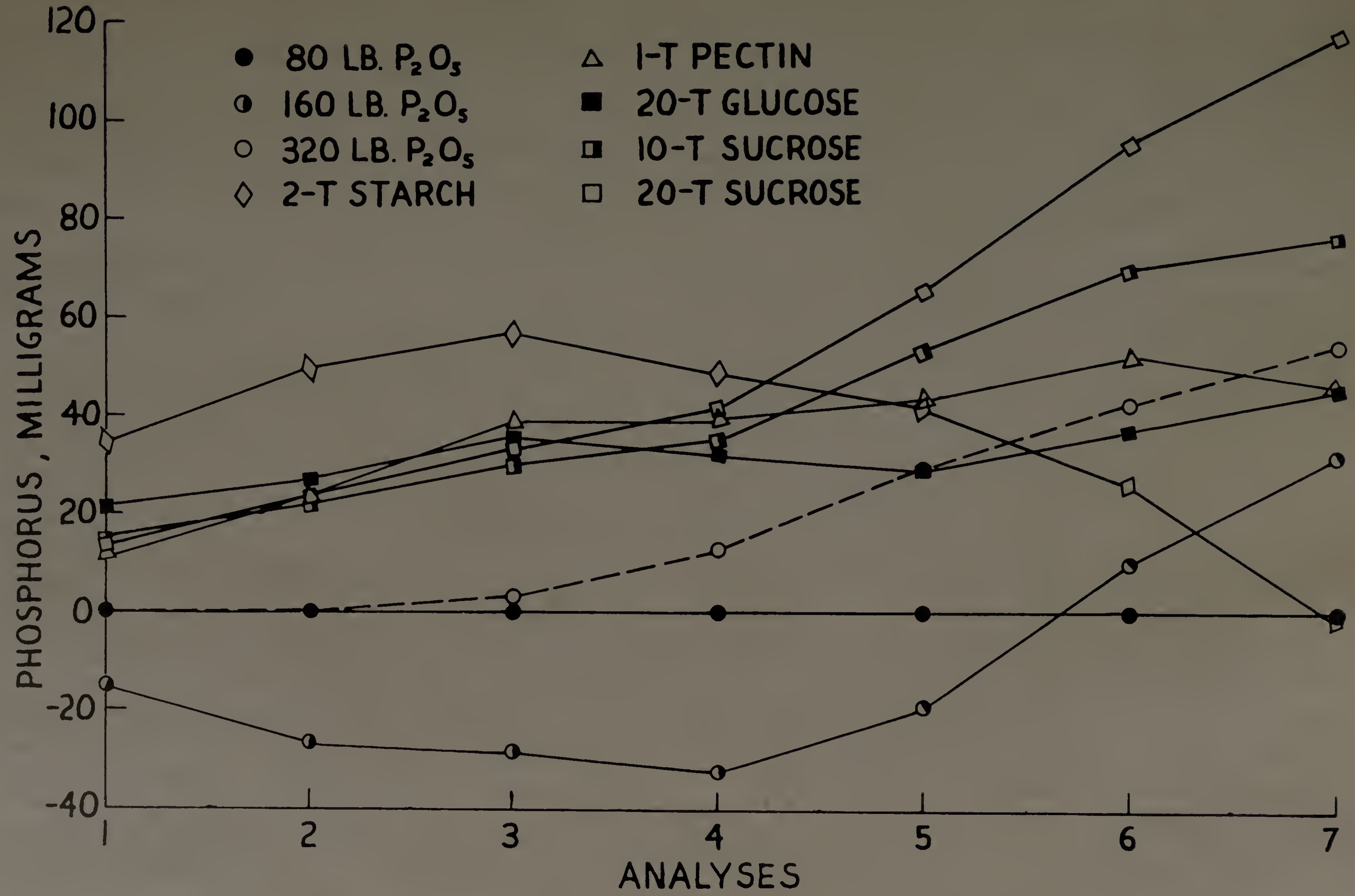

Fig. 6. Relative Uptake of Phosphates From Soils Receiving Three Different Applications of Rock Phosphate and Five Different Kinds and Rates of Organic Matter

The data show that 20 tons of sucrose were the most effective and 10 tons of sucrose, pectin, glucose, and starch were less effective.

The curves in Figure 6 show the cumulative phosphate uptake in relation to that of the 80 pounds P_2O_5 per acre as the residual phosphate source. These curves show the lack of control on the greenhouse study and that the microorganisms and their decomposition products may be effected by introducing additional variables. For no apparent reason the 160 pounds P_2O_5 gave a lower uptake than the 80 pounds until the fifth cutting. The uptake, however, was great enough for the other cuttings to give a greater total uptake. The 320 pounds P_2O_5 treatment was about equal to the 80 pounds P_2O_5 treatment until the fourth cutting and thereafter it showed a marked increase. The organic treatments pectin, glucose, and sucrose showed a cumulative increase throughout the period of study. Starch showed a cumulative decrease starting with the fourth cutting.

In Figure 4b is shown graphically the relative effects of the organic compounds with a rock phosphate residue. With the rock phosphate residue the least effective organic compound was starch. It was, however, as effective as the residual 80 pounds of P_2O_5 per acre as KH_2PO_4. The 320 pounds P_2O_5 treatment was as effective as 160 pounds P_2O_5 in the residual form of KH_2PO_4. The pectin and glucose in combination with 80 pounds P_2O_5 rock phosphate residue were as effective as the 160 pounds P_2O_5 as soluble KH_2PO_4. Both sucrose treatments with 80 pounds P_2O_5 excelled the results from the 160 pounds P_2O_5 in the soluble form.

Bauer (1) noted toxic effects on corn grown in sand cultures where organic matter had been used in combination with rock phosphate. In this study the starch alone in the soil and starch in combination with a rock phosphate residue showed different effects upon phosphate removal. The biological activity in the soil is extremely unpredictable. The end products of fermentation are very unpredictable and varied depending upon the oxygen supply of the soil. This depends to a large degree upon the moisture supply of the soil which is humanly impossible to keep constant in pot culture work. A search of the literature produced no additional information concerning the effect of rock phosphate upon the respective microorganism population which specifically decomposes starches. When causes for experimental error are considered it seems unlikely that lack of moisture control would cause so great a difference throughout a period of 42 weeks; therefore, further study of this combination seems highly desirable.

A study of the data in Figure 4b indicated that the soil responded to 80 and 160 pounds P_2O_5 per acre. They indicated that starch, glucose, and pectin increased the phosphate removed from the soil. They indicated that the soil responded from the residual effect of rock phosphate and that sucrose, glucose, and pectin resulted in the liberation of phosphates from rock phosphate. The glucose and sucrose contained no phosphorus. The starch and pectin contain phosphorus so that it amounted to the rate of 0.5 pound P_2O_5 per acre which is not significant. It seems highly reasonable to believe that the various organic acids that were liberated upon the decomposition of the organic compounds made soluble the soil phosphates and phosphates in the rock phosphate.

A more representative picture of the treatments may be realized by observing the total removal of phosphates by both corn and ladino clover. The period covered more than a year with plants growing in the soil under greenhouse conditions. The soil from replicates A and B used in the corn study was used for the ladino clover study. The phosphate removed by corn from the soil was added to that removed by the ladino clover and recorded in Tables 9 and 10 and shown in Figure 7. Starch, sucrose, glucose, and pectin gave significant increases in the soil phosphate uptake. These data seem to substantiate the fact that the organic acids that are produced by organic matter decomposition make soluble the soil phosphates and the phosphates in rock phosphate.

Influence of organic matter upon the uptake of phosphates by tomatoes. Tomato plants were used to show the influence of the organic compounds starch and citric acid upon the uptake of phosphates. The various phosphates studied were soil phosphates, KH_2PO_4 added to soil, aluminum phosphate, and iron phosphate. Swenson, Cole, and Sieling (37) worked with solutions of iron and aluminum and found that phosphorus was fixed by those cations in the form of basic phosphates. They represented these compounds by the formulas $Al(H_2O)_3(OH)_2H_2PO_4$ and $Fe(H_2O)_3(OH)_2H_2PO_4$.

Table 9. Total phosphates removed by corn and ladino clover from the soil with KH_2PO_4 source

No. of Treatment	Treatment Per Acre		Total Removal
	P_2O_5	Organic Matter	
	lbs.	tons	mgm. P
I			792.50
II	80		889.14
III	160		976.83
VIII		6-Starch	1000.79
IX	80	6-Starch	970.91
X		4-Pectin	837.39
XI	80	4-Pectin	869.40
XXIII		22-Glucose	963.91
XXIV	80	22-Glucose	924.50
XXVII			1137.90
IV		11-Sucrose	1079.20
V	80	11-Sucrose	1144.63
VI		22-Sucrose	1145.41
VII	80	22-Sucrose	1058.75

Table 10. Total phosphates removed by corn and ladino clover from the soil with rock phosphate source

No. of Treatment	Treatment Per Acre		Total Removal
	P_2O_5	Organic Matter	
	lbs.	tons	mgm. P
XII	80		839.61
XIII	160		890.01
XIV	320		921.30
XV	80	11-Sucrose	954.92
XVI	160	11-Sucrose	895.23
XVII	80	22-Sucrose	1018.07
XVIII	160	22-Sucrose	995.99
XIX	80	4-Starch	888.24
XX	160	4-Starch	856.95
XXI	80	2-Pectin	920.04
XXII	160	2-Pectin	879.17
XXV	80	22-Glucose	964.97
XXVI	160	22-Glucose	933.41

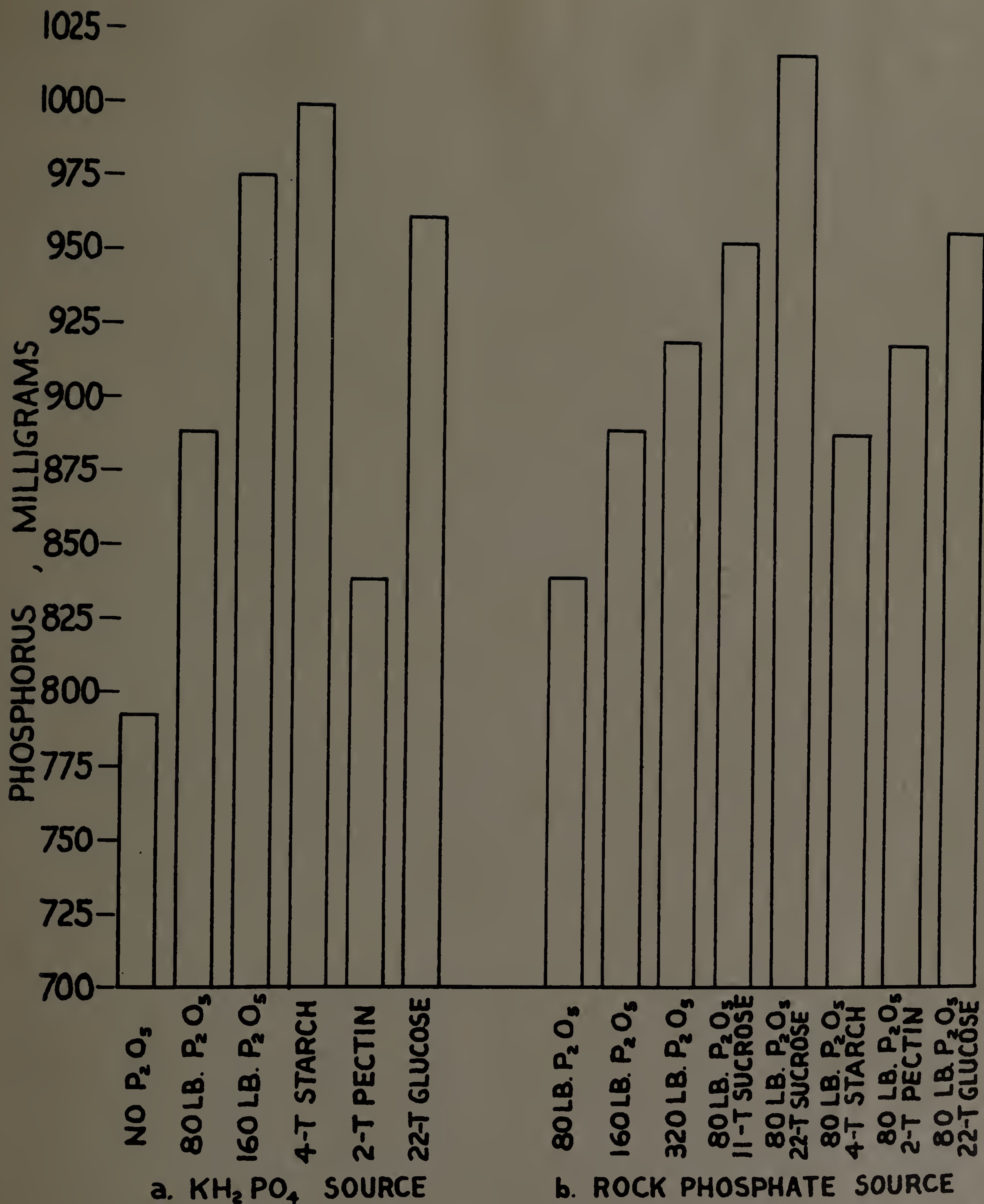

Fig. 7. Total Phosphates Removed by Corn and Ladino Clover

Russell (32) prepared freshly precipitated basic iron and aluminum phosphates at a pH of 5.5. The unused precipitates remained in the laboratory for more than a year and were thoroughly aged. Those were the iron and aluminum phosphates used in this study.

Breazeale and McGeorge (4) used the tomato plant to advantage in determining the wilting percentage of soil. Their system consisted of a container of soil surrounding the tomato plant just above the surface of the soil in which the plant was grown. This method is based on the fact that the tomato will develop roots where a portion of its stalk is in contact with moist soil. It seemed advisable to develop a similar means of studying soil chemistry by this method. This allowed the variable treatments to be in the soil in contact with roots of the plant and at the same time not to be influenced by the non-variable effects which were added to the container which held the plant.

Preliminary studies were made on the solubility of the iron and aluminum phosphates to be used. These air dry compounds were ground with mortar and pestle. To determine the total phosphorus content two 0.1 g samples were used. Each sample was dissolved in 7 ml 60 per cent $HClO_4$ and 5 ml of distilled water by heating on a hot plate. The solution was made to 250 ml, a 25 ml aliquot was taken for phosphorus determination by the method of Sherman (33). The data in Table 11 show the actual and theoretical phosphate content of the iron and aluminum phosphates.

Table 11. The phosphorus content and solubility of the iron and aluminum phosphates in water and citric acid

Formula	Percentage P		Relative Solubility in		
	Theoretical	Found	Water	0.5M Citric Acid	1% Citric Acid
$Al(H_2O)_3(OH)_2H_2PO_4$	14.62	15.15	2.4	100.0	88.8
$Fe(H_2O)_3(OH)_2H_2PO_4$	12.70	11.70	2.9	100.0	47.4

The solubility was determined for these compounds by use of water, 0.5M citric acid and 1 per cent solution citric acid. Two 0.1 g samples of each were placed into tall 250 ml beakers and 100 ml water added. They were allowed to stand for 60 minutes with occasional stirring. The solution was passed through Whatman 41-H filter paper and the residue washed three times with distilled water. The solution was made to 250 ml volume and an aliquot taken for phosphate determination. Bass and Sieling (2) used a 0.5M citric acid solution for extracting soil phosphates in determination of the fixing capacity of a soil. Two 0.1 g samples were placed into tall 250 ml beakers and 100 ml of 0.5M citric acid solution. This was placed in a boiling water bath for exactly 60 minutes. In that time both aluminum and iron compounds were completely dissolved. Dyer (9) used a 1 per cent citric acid solution to determine the probably available "mineral" plant nutrients in soils. In the present study two 0.1 g samples of each compound were placed into tall 250 ml beakers and 100 ml of citric acid solution were added. This was allowed to stand for 60 minutes and was stirred occasionally. The solution was passed through a Whatman 41-H filter paper into a 250 ml volumetric flask. The residue was washed three times with water. The solution was brought to volume and an aliquot was taken for phosphate determination. The solubility results were recorded and appear in Table 11. The compounds were completely soluble by the Bass and Sieling (2) method using a 0.5M citric acid solution. The aluminum compound appeared to be less water soluble but much more soluble in 1 per cent citric acid than was the iron compound.

To each 3 gallon, glazed jar was added 25 pounds of Merrimac fine sandy loam with a pH 5.2. To give a banding effect of the nutrient elements they were added in solution about four inches from the surface of the soil. The last four inches of soil were added after fertilization. In Table 12 the specific treatments were recorded. Nitrogen was added at the rate of 75 pounds per acre and calcium at the rate of 107 pounds per acre as $Ca(NO_3)_2 \cdot 4H_2O$. Magnesium was added at the rate of 64 pounds per acre and sulfur at 83 pounds per acre in the form of $MgSO_4 \cdot 7H_2O$. KCl was added to give 100 pounds per acre K_2O. Nitrogen and calcium were added at weekly intervals throughout the study at one-third the above rates. The treatments to the soil in the pots were identical for each.

A single tomato plant of Stockdale variety was planted in each jar. The lower leaves were removed with a razor blade up to an approximate height of 12 inches as the plant grew. The

Table 12. Treatments of nutrient elements and organic matter in the tomato study

No. of Treatment	Amount of constituents added (grams)									
	To 25 lbs. soil in pot			To rubber sleeve						
				448.3 g soil		700 g sand				
	$Ca(NO_3)_2 \cdot 4H_2O$	KCl	$MgSO_4 \cdot 7H_2O$	KH_2PO_4	Corn Starch	KH_2PO_4	Corn Starch	Al/P	Fe/P	Citric Acid
1	9.138	1.899	3.432							
2	9.138	1.899	3.432		0.897					
3	9.138	1.899	3.432	0.860						
4	9.138	1.899	3.432	0.860	0.897					
5	9.138	1.899	3.432							
6	9.138	1.899	3.432				1.4002			
7	9.138	1.899	3.432			0.1343				
8	9.138	1.899	3.432			0.1343	1.4002			
9	9.138	1.899	3.432					0.2017		
10	9.138	1.899	3.432				1.4002	0.2017		
11	9.138	1.899	3.432						0.2596	
12	9.138	1.899	3.432				1.4002		0.2596	
13	9.138	1.899	3.432					0.2017		0.2522
14	9.138	1.899	3.432						0.2596	0.2522
15	9.138	1.899	3.432			0.1343				0.2522
*16	9.138	1.899	3.432							

*No rubber sleeve.

plants were supported vertically by twine attached to 2" x 2" cross pieces in the top of the greenhouse. A rubber sleeve 7 1/2 inches long and 2 3/4 inches in diameter was brought down over the tomato plants as shown in Figure 8. A soft rubber disc of 3" diameter was made to encircle the tomato plant which passed through a 1/2 inch hole in the center. This was allowed by a slit extending from the hole in the center to the outside edge of the disc. The disc was spread apart, wrapped around the plant and closed again into the circular form. The bottom part of the rubber sleeve was stretched over the circular disc. Inside the rubber sheath the soil and/or sand containing the treatments was added after the treatments were applied.

The check treatments for the soil were with no rubber sleeve on the plant, a rubber sleeve with 448 g of soil on the oven dry basis, a rubber sleeve with 448 g of soil and KH_2PO_4 at the rate of 200 pounds P_2O_5 per acre. The variable treatments for the soil were the rubber sleeve containing 448 g of soil with corn starch at the rate of 2 tons per acre, rubber sleeve containing 448 g of soil with corn starch at the rate of 2 tons per acre and KH_2PO_4 at the rate of 200 pounds of P_2O_5 per acre.

For the other part of this experiment washed beach sand was used. The beach sand was passed through a 1 mm sieve. A 7 per cent solution of HCl was allowed to stand on the sand for seven days. The HCl solution was removed and the sand washed in a churn with distilled water until the solution coming off

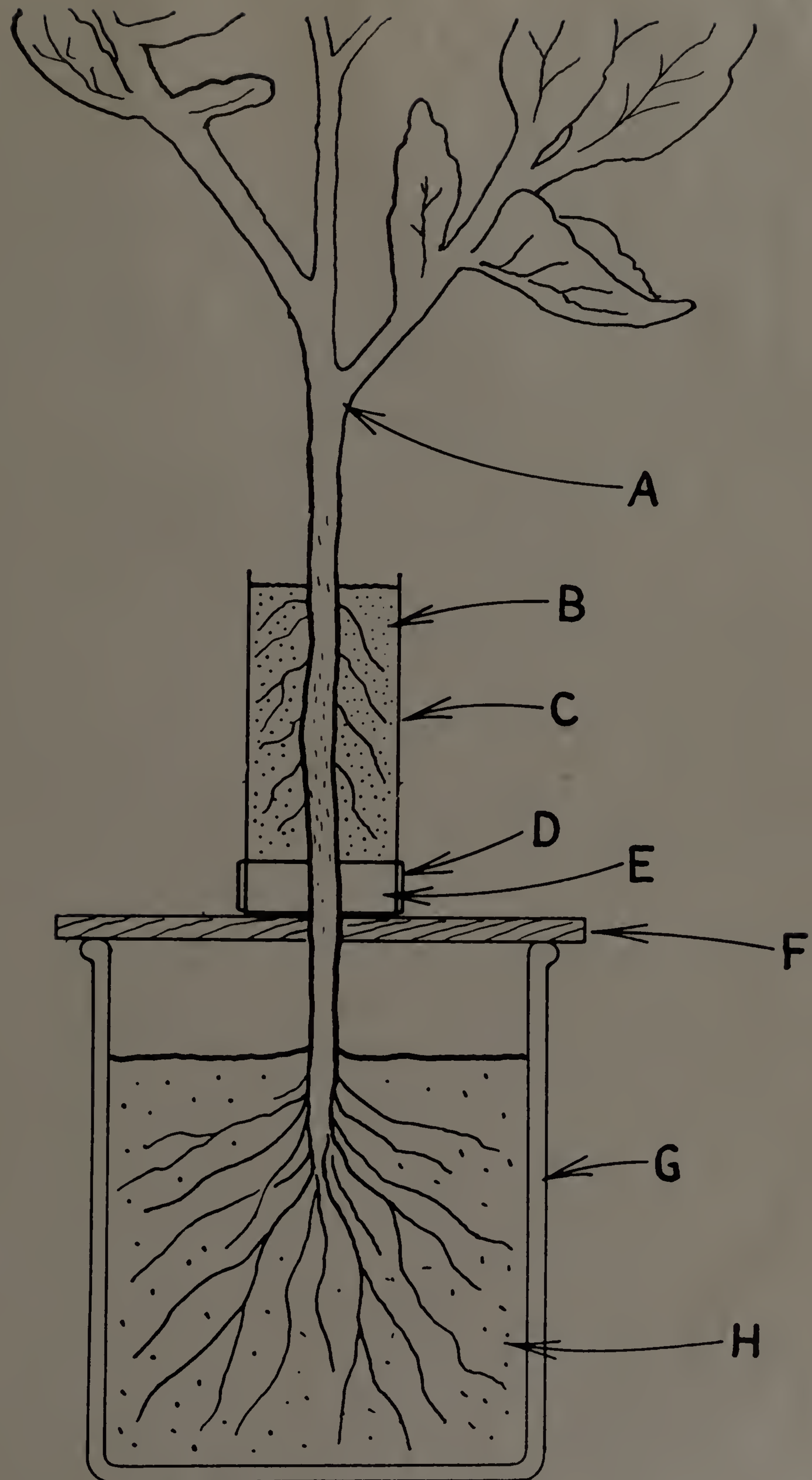

Fig. 8. Method of Determining Phosphate Uptake From Various Sources and as Influenced by Organic Matter

A, tomato plant; B, soil or sand; C, rubber sleeve; D, elastic band; E, sponge rubber base; F, wooden support; G, container; H, soil.

was pH 5.7 which was approximately that of the distilled water. The sand was then dried to the air dry condition.

The check treatments for the sand were the rubber sleeve containing 700 g of sand on the air dry basis, rubber sleeve containing 700 g of sand and KH_2PO_4 at the rate of 200 pounds of P_2O_5 per acre, rubber sleeve containing 700 g of sand and alumino-phosphate at the rate of 200 pounds of P_2O_5 per acre, rubber sleeve containing 700 g of sand and iron-phosphate at the rate of 200 pounds of P_2O_5 per acre. The variable treatments were added to the rubber sleeve containing 700 g of sand. The variables were corn starch at the rate of 2 tons per acre, corn starch at the rate of 2 tons per acre and KH_2PO_4 at the rate of 200 pounds P_2O_5 per acre, corn starch at the rate of 2 tons per acre and alumino-phosphate at the rate of 200 pounds P_2O_5 per acre, corn starch at the rate of 2 tons per acre and iron-phosphate at the rate of 200 pounds P_2O_5 per acre, alumino-phosphate at the rate of 200 pounds P_2O_5 per acre and an equal milliequivalent weight of citric acid, iron-phosphate at the rate of 200 pounds P_2O_5 per acre and an equal milliequivalent weight of citric acid, KH_2PO_4 at the rate of 200 pounds P_2O_5 per acre and an equal milliequivalent weight of citric acid.

The tomato plants were transplanted May 22, 1950 and harvested September 5, 1950 without being allowed to bear fruit. The treated soil and sand treatments were added to the respective rubber sleeve and the $Ca(NO_3)_2 \cdot 4H_2O$ in solution added weekly to the jars from July 29 through August 28, 1950. The content of both the pots and rubber sleeves was kept supplied with moisture throughout the study.

An unavoidable delay in the arrival of the rubber supply hampered the study. The plants were almost through their rapid growing period before the soil was added to the rubber sleeves. That meant that by the time the roots had developed in the rubber sleeve the plant was almost mature. Before the plants stopped growing a treatment of cyanogas applied to the greenhouse killed some of the tip leaves of the tomatoes. After growing approximately an additional foot tall the tips died altogether. This happened to a large number of plants being studied and thus terminated the study prematurely.

To harvest and analyze the total plant would have diluted the phosphorus in the total sample. This was alleviated by harvesting each plant in two sections. The top section was taken 12 inches from the growing tip. The body section was taken 8 inches from the bottom of the rubber sleeve. The plant material was oven dried, ground in a Wiley mill, and two 1 gram samples were taken from each section for determining the phosphorus content.

The yield and phosphate removed by the tomato plants of 3 replicates were recorded in Table 13. These data show positive results only in cases where KH_2PO_4 alone was added to sand. There was no apparent benefit where either starch or citric acid was added. KH_2PO_4 showed no significant response where added to soil alone. There is slight evidence that starch was

Table 13. Treatments of nutrient elements and organic matter in the tomato study

No. of Treat-ment	Constituents added to rubber sleeve: SOIL lbs. per acre P_2O_5 KH_2PO_4	SOIL Starch	SAND lbs. per acre P_2O_5 KH_2PO_4	SAND P_2O_5 Al/P	SAND P_2O_5 Fe/P	SAND Starch	SAND Citric Acid	Removal: Plant material gm.	Phosphorus mgm.	%P Tops	%P Body
1								155.39	313.32	0.300	0.190
2		4000						169.44	319.18	0.350	0.177
3	200							161.98	307.99	0.304	0.178
4	200	4000						163.82	304.24	0.340	0.173
5								157.67	313.27	0.309	0.186
6						4000		171.38	338.41	0.309	0.187
7			200					166.70	342.03	0.291	0.196
8			200			4000		164.45	326.77	0.321	0.188
9				200				153.92	309.23	0.344	0.190
10				200		4000		165.38	330.07	0.316	0.190
11					200			146.15	314.25	0.340	0.206
12					200	4000		159.62	319.60	0.349	0.189
13				200			720	150.00	312.77	0.387	0.196
14					200		720	162.05	318.38	0.304	0.184
15			200				720	163.62	338.00	0.309	0.195
*16								155.35	309.03	0.339	0.186

*No rubber sleeve.

beneficial when used in combination with the $Al(H_2O)_3(OH)_2H_2PO_4$ and the $Fe(H_2O)_3(OH)_2H_2PO_4$. The sectional analyses of the plants proved to be of value in that a greater concentration of phosphate was found to be in the top portion of the plants. In Table 13 the data for the sectional analyses of the plants show that there was a greater concentration of phosphate in the top and that the variation covered a wider range than was shown in the remaining part of the plant. In a study of this type it seems desirable to make sectional analyses of the plants which will show greater differences between treatments.

It is believed that this technique has an important place in the study of soil chemistry. This study exposed many of the pitfalls of the technique as many unforeseen problems arose. It is suggested that the rubber sleeve be placed around the plant and the treated sand and/or soil added when at most 3 or 4 leaves are beyond the top of the sleeve. The leaves below should be removed. The sleeve should be supported approximately 1 inch above the surface of the medium used. Sufficient nutrient elements should be added to force the plants from this time until harvested. It appeared that the greatest concentration of phosphorus was in the tops. A sectional analysis of the plant seems advisable in future studies.

DISCUSSION

Results of this study indicated that the phosphates that have become fixed in the soil may be released by organic acids produced in the soil. One realizes that nature has provided a natural means for supplying phosphates to plants. That is realized in forest soils where apparently no available phosphates are added. Those phosphates added by the return of leaves to the soil would be expected to be fixed in the soil before a depth of 2 or 3 inches is penetrated. By now one must realize that the microorganisms play their role in decomposing the organic matter and producing organic acids that release the fixed phosphates.

Polyuronides and uronic units are widely distributed in nature as products of plants, animals, and microorganisms. It seems reasonable to believe that the uronic acid group plays a part in the phosphate release. Norman and others (10,11,12,14, 15,16,25,26,27,34,39) have studied the polyuronides and it has been stated that as much as 10 to 38 per cent of the organic carbon in soils is uronic.

Citric acid is produced when fungi decompose organic matter. *Aspergillus niger* has given successful results in producing citric acid on an industrial basis according to Prescott and Dunn (31). It seems logical to expect citric acid to be formed in small amounts in acid soils with these fungi always present.

Fraser (14) pointed out that in moderately dry soil over

30 per cent of the organic matter may be microorganisms--living and dead. He believed that further progress in the study of soil organic matter is more likely to be made from the micro-biochemical side than from application of direct chemical analyses to soil organic matter in the mass. Hester (18) pointed out that in pot cultures very little response was obtained from the addition of phosphates where the organic matter content was 3 per cent.

The present study showed that rock phosphate may have an important role in agriculture where proper management is a practice. Data in this study indicate that for quick response the available form of phosphates is desired. They show that response was realized from rock phosphate after a period of time. Rock phosphate, however, should not be considered a substitute for the more available forms of phosphates but as a supplement for them. The P_2O_5 in the rock is less expensive to add pound for pound.

Brown (5) showed that biennial surface applications of superphosphate in acid soils penetrated no more than 2 or 3 inches and perhaps less in 16 years, whereas, rock phosphate similarly applied penetrated 7 inches. It is not surprising that Bauer's work (1) failed to indicate a solvent action of decaying organic matter on rock phosphate. He moistened the soil and organic matter and let it set in the greenhouse for 5 months, then let the soil become air dry and then tested for soluble phosphates. Evidently the sesquoxides became hydrated and tended to increase the phosphate fixing power of the soil.

He did show, however, that in some cases mixtures of organic matter and rock phosphate increased the phosphate uptake. That investigator failed to appreciate the fact that the phosphate release is a slow but steady process and the plant roots must be there to utilize those released phosphates as they are liberated by the organic acids. Paul and DeLong (28) showed that prolonged flooding of soils followed by desiccation to air dry state under aerobic conditions produced undesirable effects with respect to the solubility relations of both native soil phosphate and added water soluble phosphates. They gave the acids credit for changing the pH of the solution but did not give them credit for the downward movement of the phosphorus in the soil which they noted especially in the soil-glucose solution system.

SUMMARY

Organic compounds α d-galacturonic acid, citric acid and citrus pectin in a 0.2N NaOH solution were found to be effective in preventing phosphate precipitation by iron. The dl-phenylalanine showed a negative effect in preventing phosphate precipitation by iron. The effectiveness of citric acid was found to vary according to the sequence of adding the anions and iron which indicated a need for a standard procedure for this study.

Pot cultures of corn were used to study the effect of organic matter which contained no phosphorus on the soil phosphates and rock phosphate. The soil phosphates responded to soluble phosphates added to the soil. Sucrose added at the rate of 2 tons per acre showed a 21 per cent increase in soil phosphate removed. Starch and pectin added alone to the soil gave no response by the corn. The corn showed no significant response to rock phosphate alone. Corn did respond to rock phosphate in combination with sucrose, starch, glucose, and pectin.

Pot cultures of ladino clover were used to show the effect of organic compounds on residual KH_2PO_4 and rock phosphate that served as variable treatments for the above corn study. The clover showed a response to the residual KH_2PO_4. The soil phosphates responded to the addition to the soil of starch, glucose, and pectin. Starch and glucose caused a greater uptake of phosphates from the soil than did the residual effect of the KH_2PO_4. The soil with the residual rock phosphate showed in each case that the addition of organic matter resulted in an increase in

phosphates removed. Indications were that microbiological activity in the soil produces small amounts of organic acids which release to the plant the soil phosphates and phosphates from rock phosphate.

A new method for studying soil chemistry was investigated. The variable treatments were made in a rubber sleeve which contained soil or sand and in which tomato plants had been allowed to establish adventitious roots. The plants were forced by adding the check treatments to the soil that held the plants. This technique showed favorable possibilities.

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